The Healthy Adrenal

Introduction

This first lesson on the adrenal gland will serve as an overview and introduction to the entire gland. We will introduce the gross anatomy, histologic layers and their embryonic origins, cells of each layer, and hormones they produce. We will discuss the broad strokes of the endocrine feedback systems that regulate each layer, but only at a high level—the mechanisms of the individual receptors and their cytoplasmic targets will be discussed in the subsequent lessons specific to each hormone. Finally, we'll close with a discussion of how the hormones overlap—either in their production and release or their intended metabolic effect—saving intense discussion on their uniqueness for the lessons dedicated to each hormone.

Gross Anatomy and Vasculature

The **adrenal glands** (also known as **suprarenal glands**) are a pair of organs, each located on top of a kidney. Their name describes their location relative to the kidney (ad = near, supra = above), but fails to communicate what the gland actually does. The gland can be divided into two embryologically distinct regions—cortex and medulla. The **cortex** can be further subdivided into three zones. Each zone is responsible for making a different hormone. Each hormone of the cortex is a **steroid hormone** derived from **cholesterol**. The cortex is derived from **mesoderm**. The **medulla** is the catecholamine-secreting zone. It lies deep in the gland, forming the center of the gland. The medulla is derived from **neural crest cells**. The gland is also surrounded by a **capsule.**

The vasculature that perfuses these glands atop the kidneys is named the **suprarenal arteries**. There is a superior, middle, and inferior suprarenal artery. The gland is drained by **suprarenal veins**. On the left side, which is farther from the inferior vena cava, the suprarenal vein, gonadal vein, and renal vein coalesce to cross the midline and drain into the IVC. On the right, closer to the inferior vena cava, those veins each drain into the IVC directly.

The arteries penetrate the gland from the capsule and work their way toward the middle. **Capsular branches** of the suprarenal arteries provide the gland with a unique blood supply. The capsular branches provide both **cortical arterioles** that supply the cortex with oxygenated blood and **medullary arterioles** that bypass the cells of the cortex and bring oxygenated blood directly to medullary cells. All blood flows from the outer cortex (where the capsule is) toward the center of the gland. Venules become small veins, and all veins eventually drain into the **central adrenomedullary vein**. Both the cortical and medullary arterioles open into a **fenestrated capillary bed**. The cortical capillaries are confluent with the medullary capillary beds. This means that the **medulla has a dual blood supply**—oxygenated blood from the medullary arterioles and deoxygenated blood rich in the hormones secreted by the cortex. The central adrenomedullary vein and its tributaries are unique veins in that they have a very conspicuous, very large, and very strong **tunica media**. These vessels are rich in smooth muscle, and their synchronized contraction can serve to drain the adrenal gland of volume, shrinking its size and increasing the output of catecholamines from the medulla—an action akin to wringing out a sponge.

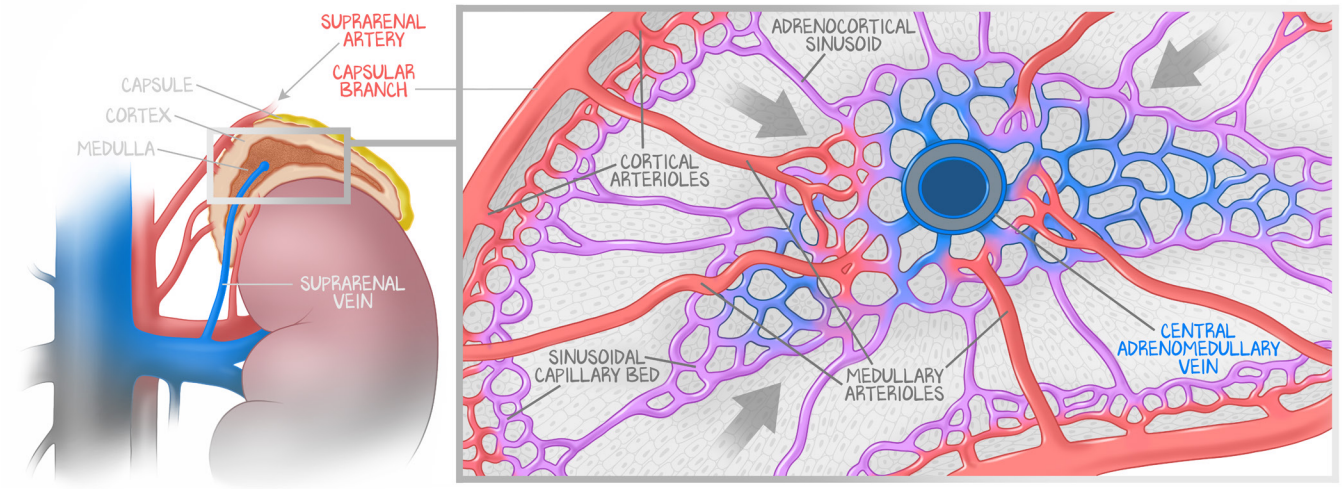

Figure 1.1: Adrenal Gland Vasculature
The adrenal gland is the shape that it is because gravity pulls it down against the kidney. There is no functional top or bottom to the gland. All the vessels originate from capsular arteries along the outside of the gland and penetrate inward toward middle veins. From the capsular arteries arise cortical arterioles that irrigate the cortex with oxygenated blood, and medullary arterioles that bypass the cortex to deliver oxygenated blood to the medulla. In addition, cortical capillaries deliver a second vascular supply to the medulla, which is deprived of oxygen but rich in cholesterol hormones.

Zonation of the Cortex—The Advanced Organizer

The adrenal cortex is divided into three zones based on the arrangement of its cells and what they make—the zona glomerulosa, zona fasciculata, and zona reticularis. These are scary names for new learners, and the material can quickly get out of control—if you let it. But with a simple advanced organizer and a little understanding of physiology, you can ensure you never forget. The cortex is regulated by the anterior pituitary and the RAAS.

The adrenal gland is a fat hat, sitting on the kidney. In the kidney, the main thing we cared about was filtration. Filtration is measured by the **glomerular filtration rate**. That would be, in order, G then F then R. This helps remind you of the order of the layers, from the outside of the gland toward the medulla—G**lomerulosa** is the most cortical, the F**asciculata** is next, and the R**eticularis** is the deepest layer of the cortex, surrounding the medulla.

"It gets better the deeper you go—salt, sugar, sex." The hormones these zones make are mineralocorticoids, glucocorticoids, and sex hormones. **Mineralocorticoids** affect the collecting duct and enhance the reabsorption of salt. **Glucocorticoids** have numerous effects, but their very obvious one is to increase blood glucose (sugar). **Adrenal androgens** are very weak sex hormones and play little role in normal sexual development, but can cause problems in excess. Regardless, they are sex hormones.

GFR + salt, sugar, sex. The glomerulosa is responsible for the synthesis of the mineralocorticoid **aldosterone**. The fasciculata is responsible for the synthesis of the glucocorticoid **cortisol**. The reticularis is responsible for synthesizing adrenal androgens, dehydroepiandrosterone (**DHEA**), and dehydroepiandrosterone sulfate (**DHEAS**).

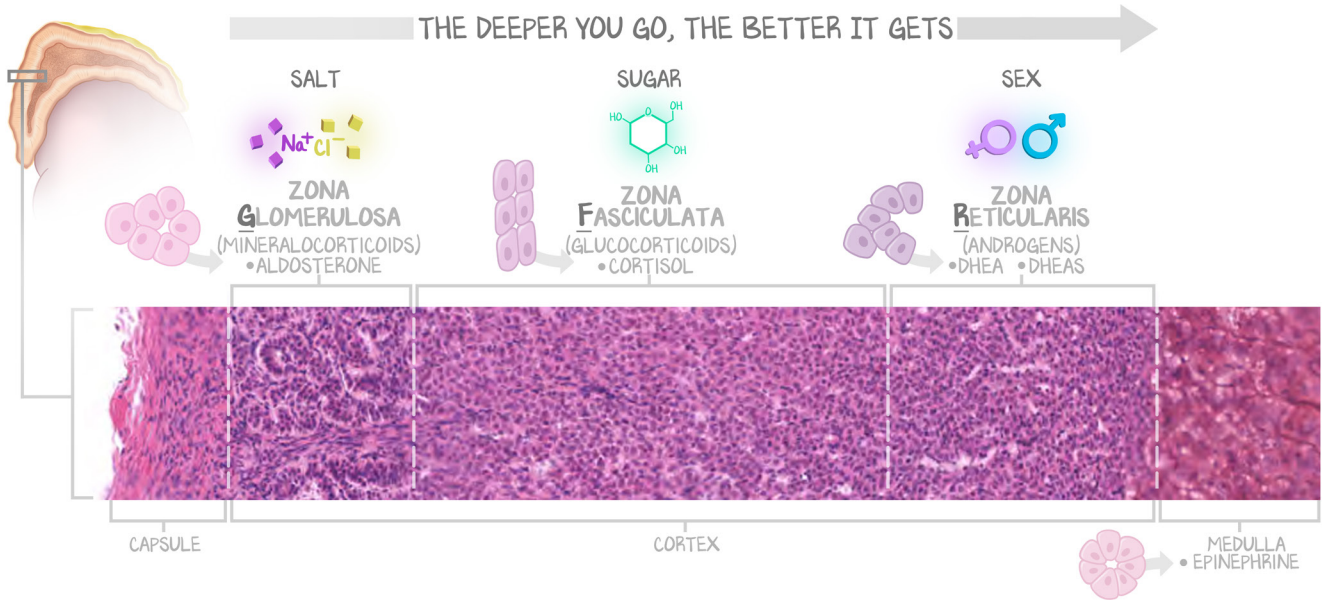

Figure 1.2: Zonation of the Adrenal Gland
From the outside in, the zona glomerulosa, fasciculata, and reticularis make aldosterone, cortisol, and androgens, respectively. That is, the glomerulosa makes salt mineralocorticoids, the fasciculata makes sugar glucocorticoids, and the reticularis makes sex hormones; "*it gets better the deeper you go*." The cells of the fasciculata and reticularis both possess the ability to make either cortisol or DHEA and so appear similarly on histology. The glomerulosa alone makes aldosterone and has a different appearance from that of the other two cortical layers. Finally, the medulla, which makes norepinephrine and epinephrine, is embryologically distinct from the cortex and appears vastly different on histology. Check out the supplementary videos for electron micrographs and histology in detail.

Histology and Physiology of the Cortex

Many texts state the vasculature of the adrenal cortex to be fenestrated, sinusoidal, or both. The only sinusoidal capillaries in the body are within hematopoietic organs—bone marrow, liver, and spleen. Just like the rest of the endocrine system, **fenestrated** capillaries (with many small pores) enable peptide hormones to exit the blood and affect the cells of the adrenal gland. Those texts may be confused or may be using sinusoidal as meaning "the form of a sine curve." Those texts are certainly confusing. Based on histology, electron microscopy, and the function of endocrine glands—peptide hormones are small and necessitate only pores (fenestrae), not sinusoids—the vessels here are **fenestrated**.

Zona glomerulosa. The zona glomerulosa comprises a thin layer of cells (~15% of the cortex) just under the capsule. It consists of clusters or curved columns of closely packed columnar cells. These columns, surrounded by fenestrated capillaries, extend through the zona fasciculata and are continuous with the zona reticularis. The zona glomerulosa is responsible for the synthesis of the mineralocorticoid **aldosterone**. Aldosterone's intended effect is in the **collecting duct** of the kidney. Aldosterone is a steroid hormone (Gen Pharm #7: *Receptors and Second Messengers*), so it will bind to a cytoplasmic mineralocorticoid receptor in the cytoplasm of target cells. There, it displaces its receptor's chaperone, heat shock protein 90 (Hsp90), and, together with the receptor, translocates to the nucleus and modifies transcription. In the **principal cells** of the collecting duct, it increases transcription of **ENaC channels.** ENaC channels (Renal: Kidney #4: *Regional Transport and Pharmacology*) increase sodium reabsorption from the collecting duct in exchange for potassium. In **intercalated cells** of the collecting duct, aldosterone increases the transcription of **carbonic anhydrase**, resulting in the accelerated wasting of H^+ into the urine and bicarbonate into the blood. Wherever ENaC channels are used, aldosterone is the signal that drives them. ENaC channels serve to conserve sodium and are located in the colon (increased absorption), sweat glands (increased reabsorption), and collecting duct (increased reabsorption).

Increasing sodium has nothing to do with the sodium level on a basic metabolic profile. Sodium on the BMP is a reflection of osmolarity, of how diluted or concentrated the blood is. Sodium reabsorption is responsible for **volume expansion**. Because "water follows salt," if the sodium is reabsorbed from the collecting duct, volume increases to balance the sodium. Because the sodium draws water to it, the result is an isotonic increase in volume. Volume expands in order to improve the perfusion pressure to the glomerulus (hence the name glomerulosa). In times of a low glomerular filtration rate, the macula densa of the JG apparatus senses low flow through the system and instructs the JG cells of the JG apparatus release renin (Renal: Kidney #3: *Glomerular Filtration*). The desired outcome of renin release is to raise the blood pressure through vasoconstriction (Ang-2) and volume expansion (aldosterone) via reabsorption of sodium at the cost of wasting potassium. Both increased blood pressure and circulating volume improve GFR. The RAAS involves the activation of **Ang-2**, Ang-2 "tensing the angios" and **stimulating aldosterone release**. Therefore, the glomerulosa is **independent of the hypothalamic-pituitary axis**. Much more on this in Adrenal #3: *Aldosterone*.

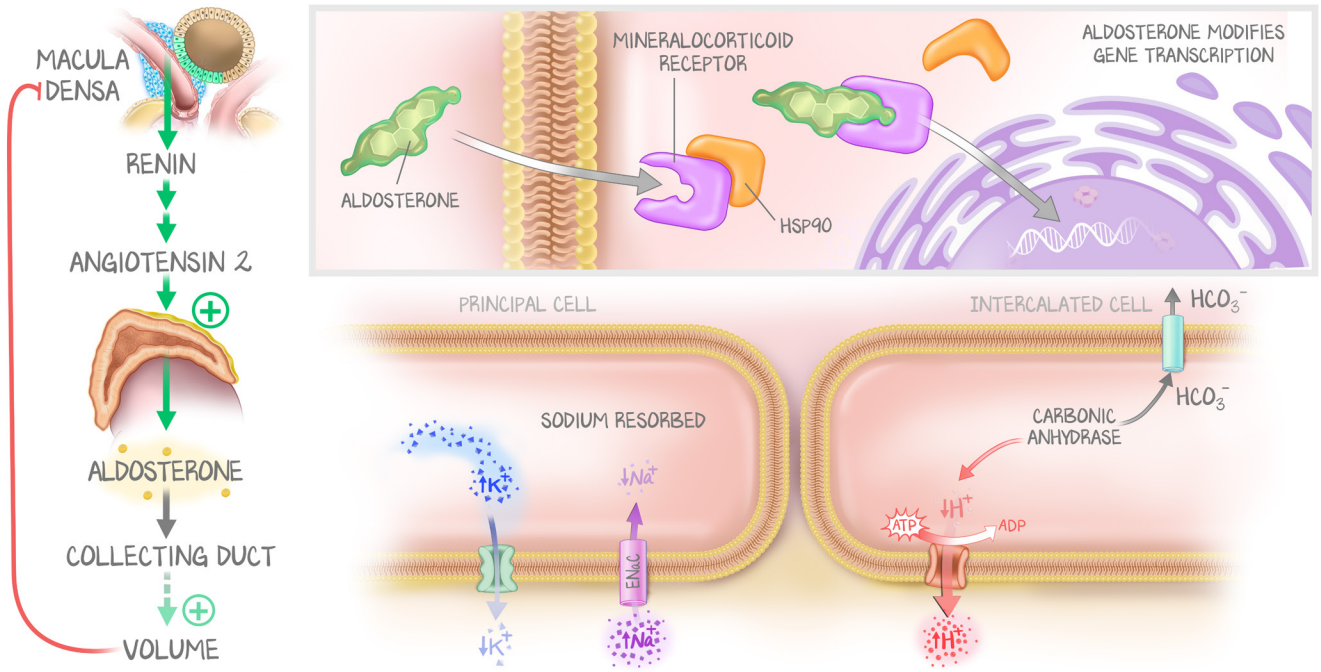

Figure 1.3: Zona Glomerulosa Regulation
This is a bit of a stretch, but we wanted to capitalize on your knowledge of the hypothalamus-pituitary axis to make this explanation easier. We have represented aldosterone regulation, the regulation of the zona glomerulosa, using the model of HPA axis regulation. See the macula densa as the hypothalamus, angiotensin-2 as the hypothalamic hormone, the adrenal gland as the anterior pituitary, aldosterone as the anterior pituitary hormone, the effector organ as the collecting duct, and the effector "hormone" as volume. Angiotensin-2 and aldosterone are the positive feedforwards down the axis, and the product of the axis, the volume, negatively feeds back on the axis. As volume increases, the GFR improves and thus silences the macula densa. To the right, we show aldosterone as a steroid hormone, displacing Hsp90 from its receptor in the cytoplasm of target cells, such as those of the collecting duct. Aldosterone then increases the transcription of ENaC channels (reabsorbing sodium) and H^+ ATPase (alkalizing the blood).

Zona fasciculata. The fasciculata takes up nearly 80% of the cortex. The cells of the zona fasciculata are huge compared to those of the glomerulosa. On histology, there appear to be elongated columns of cells flanked by fenestrated capillaries. Of course, in the three dimensions, each column—only two cells thick—is surrounded by capillaries. In a two-dimensional longitudinal section, they appear to be adjacent. Having this much vasculature for every cell ensures a large surface area for the cells to receive peptide hormone signals. The cells of the fasciculata make cortisol, a lipophilic steroid hormone. They

appear large and stain a light pink on H&E staining because of the abundant synthesis of cortisol. On electron microscopy, the cytoplasm is abundant with lipid droplets and has a highly developed smooth endoplasmic reticulum and abundant mitochondria. This mirrors the various other locations of steroid hormone synthesis (as discussed in the final section of this lesson). The principal secretion of the fasciculata is the glucocorticoid **cortisol** (although it does retain the ability to secrete adrenal androgens, you should think of this zone as secreting only cortisol). Like aldosterone, cortisol is a **steroid hormone**, has a glucocorticoid receptor chaperoned by Hsp90, and translocates to the nucleus when bound to its receptor. Cortisol has many effects, the most obvious of which are in relation to metabolism. Cortisol is one of the anti-insulin hormones, and so has the opposite effect, shifting the body into a **glucagon-dominant state** (Biochemistry: Metabolism #2: *Glucagon vs. Insulin*). The liver is shifted towards **gluconeogenesis** (fatty acid oxidation, glycogenolysis) and provided the resources to do it—**adipose** tissue **releases fatty acids,** and skeletal muscle **releases amino acids**. In nonmetabolic tissue, the utilization of glucose is reduced, and there is a shift to using glycogen and fatty acid oxidation (for the tissues that can). In **fibroblasts**, cortisol inhibits protein synthesis, so it is used in autoimmune diseases that result in scarring due to overactive fibroblast activity. Cortisol worsens immunity, so exogenous glucocorticoids are used in autoimmune disease, but endogenous cortisol merely impairs immunity to infections. Cortisol release is under the influence of the hypothalamic-pituitary axis. CRH released by the hypothalamus stimulates the release of ACTH from the anterior pituitary, which in turn stimulates the production and release of cortisol from the fasciculata. That ACTH signal both induces the production of cortisol and sustains a trophic signal. Without ACTH, the fasciculata will atrophy. The production of cortisol inhibits CRH and ACTH; the release of ACTH inhibits CRH. Much more on cortisol in Adrenal #2: *Cortisol*.

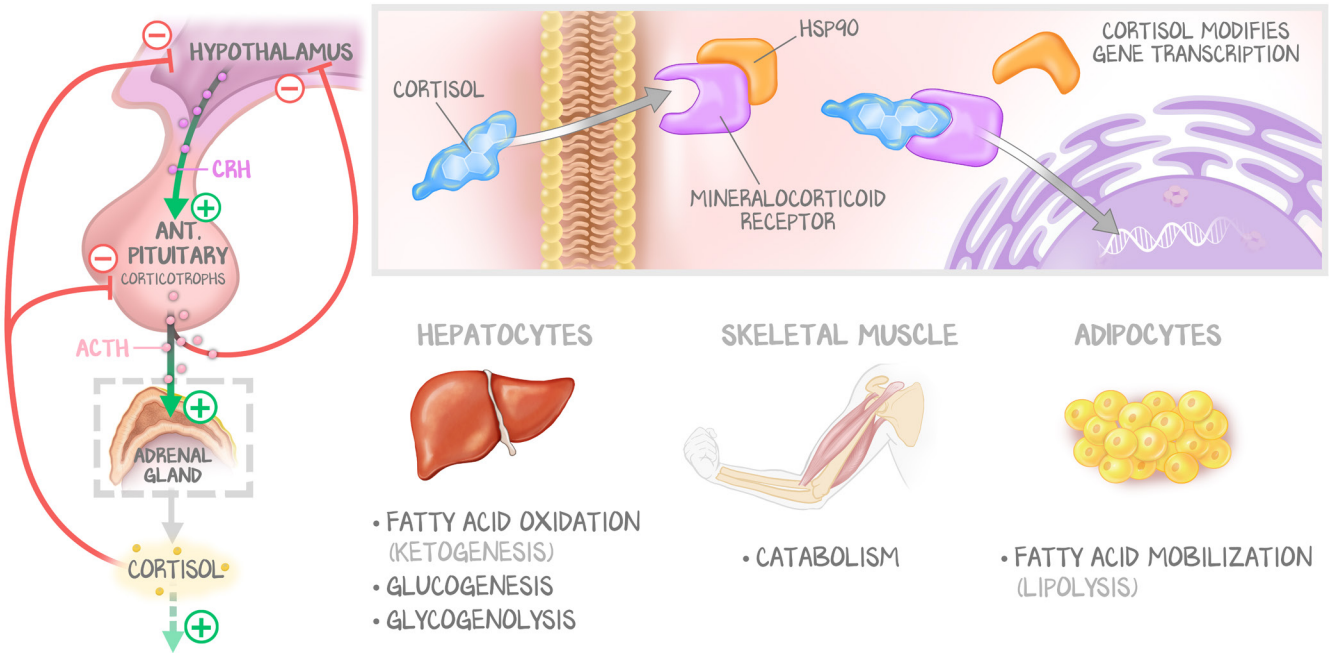

Figure 1.4: Zona Fasciculata Regulation and Effect
Under the control of the hypothalamic-pituitary axis, the zona fasciculata increases cortisol production under the influence of ACTH, released from the anterior pituitary under the influence of hypothalamic CRH. In classic negative feedback inhibition up the axis, cortisol inhibits ACTH and CRH release. Cortisol, like aldosterone, is also a steroid hormone, and therefore displaces Hsp90 and translocates to the nucleus. Cortisol affects all cells, but its effect on the cells of metabolism mirrors that of glucagon—hepatocytes make fuel for everyone else, skeletal muscle catabolizes proteins and sends amino acids to the liver, and adipocytes mobilize fatty acids back to the liver.

Zona reticularis. The zona reticularis accounts for only 5% of the adrenal cortex. Much less is known about this zona and the effects of its products—DHEA and DHEAS. DHEA and DHEAS are **much less potent** than the androgens produced by the gonads. And they are androgens, male-phenotype-inducing. These hormones are involved in the generation of secondary sex characteristics in females. Testosterone by the testes vastly overshadows the androgenic effects of DHEA and DHEAS in males. In females, DHEA and DHEAS are responsible for the growth of axillary and pubic hair. DHEA and DHEAS can also be converted to testosterone in peripheral tissues by the enzyme 17-ketosteroid reductase. Thus, in excess, DHEA and DHEAS may have virilizing (making-male-phenotype) effects. The most important thing to learn about the reticularis is that although its products, DHEA and DHEAS, are androgens, they are **not influenced by LH or FSH**, and the cells of the zona reticularis are instead **stimulated by ACTH** (just like the zona reticularis). How DHEA and DHEAS regulate the endocrine feedback loop on CRH and ACTH is uncertain. Their impact on health and disease will be covered in more detail in Adrenal #4: *Adrenal Hyperplasia NOS* and is almost always tested in relation to congenital adrenal hyperplasia.

Histology and Physiology of The Medulla

The medulla is an extension of the sympathetic nervous system. It is the truest neuroendocrine tissue in the body. The **chromaffin cells** (also called medullary cells) are **secretory** cells that release **catecholamines,** such as **epinephrine,** when the sympathetic nervous system is activated. The chromaffin cells are derived from **neural crest cells,** and the adrenal medulla represents a highly sophisticated sympathetic ganglion. The chromaffin cells receive inputs from preganglionic fibers (first-order neurons) of the sympathetic nervous system. Their output is epinephrine into circulation catecholamines. The difference is that these cells do not have axonal projections, but rather release hormones into the circulation.

Preganglionic sympathetic nerves (Gen Pharm #8: *Intro to Autonomics*) innervate the chromaffin cells. This is a first-order connection, and so these sympathetic nerves release **acetylcholine** at the synaptic cleft, activating **ionotropic acetylcholine receptors** (nicotinic AChR, also in Gen Pharm #8: *Intro to Autonomics*), which leads to chromaffin membrane depolarization. This depolarization induces calcium influx and the subsequent fusion of lysosomes with the plasma membrane. That process sounds very similar to the process experienced by postganglionic neurons of the sympathetic nervous system. But the depolarization of the postganglionic neuron releases, in turn, a neurotransmitter at a second nerve terminal at the effector organ. Chromaffin cells release their vesicles not into a nerve terminal, but instead into the **bloodstream**. Thus, they are endocrine cells because they release hormones into the blood, but they are neural cells because they are innervated by first-order sympathetic fibers.

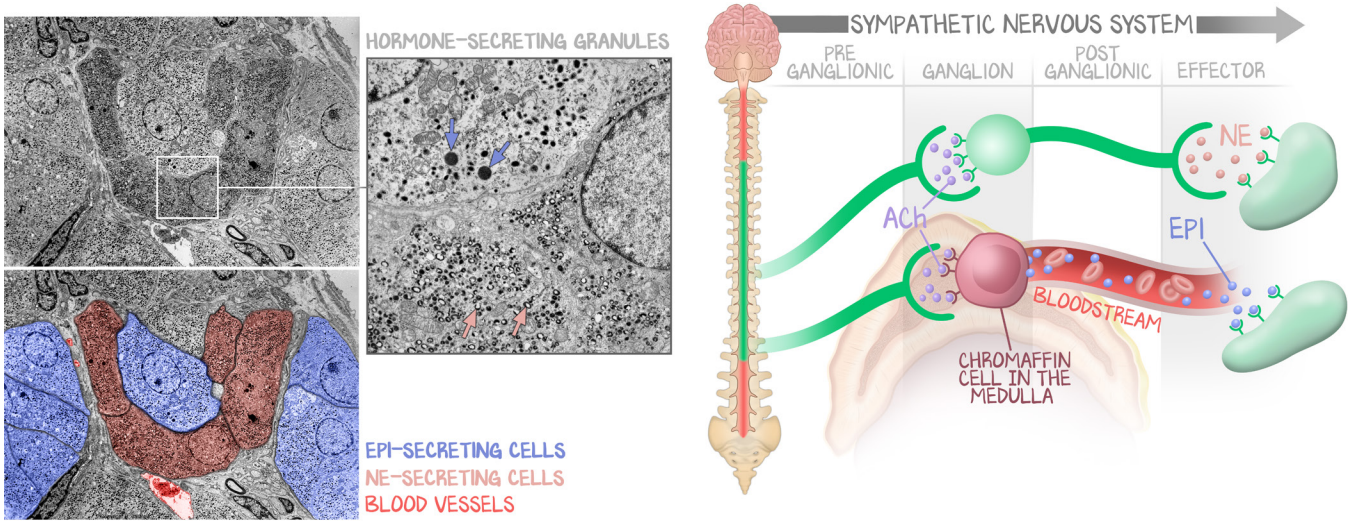

Figure 1.5: Chromaffin Cells of the Medulla
(a) Electron micrograph showing two types of chromaffin cells. The norepinephrine-secreting cells are identified by their darker cytoplasm and black and white granules. The epinephrine-secreting cells possess purely black vesicles (can contain a rim of grey) and lighter cytoplasm. (b) A visualization of the similarities between the chromaffin cells; the release of epinephrine into the bloodstream is analogous to the release of norepinephrine by the second-order sympathetic fiber's axon.

In experimental models, chromaffin cells **grow axonal processes** when in isolation. In the same experimental model, the addition of glucocorticoids **prevents the development of axonal processes**. Glucocorticoids are also responsible for the transcription of the enzyme that converts **norepinephrine to epinephrine**. The default is for a chromaffin cell to synthesize only norepinephrine and grow an axon, just like a second-order sympathetic neuron, and just like neural crests would do in a ganglion. The blood flow from the cortex that drains through the medullary capillaries on its way to the central vein ensures a constant supply of glucocorticoids to the medulla. The presence of glucocorticoids maintains the chromaffin cells and enables their production of epinephrine from norepinephrine. Any norepinephrine released from the medulla is short-lived and has limited impact circulating through the blood. Any norepinephrine released is likely a product of insufficient induction of epinephrine-making enzymes.

Embryology and the Fetal Adrenal Gland

The **medulla** is derived from **neural crest** cells. The medulla is derived from the sympathetic ganglion and is an extension of the sympathetic nervous system. The **cortex** is derived from **mesoderm**. "Cortex" is more than just cortex during development. Use Figure 1.6, below, to follow along with the text.

The **fetal cortex** and the **permanent cortex** are two distinct structures. The fetal cortex serves the role of the cortex—synthesizing hormones, being ACTH responsive—for almost the entirety of gestation. After birth, the permanent cortex takes over, and the fetal cortex disappears. There is a gradual shift from the fetal cortex to the permanent cortex, with accelerated involution of the fetal cortex at birth.

Initially, the chromaffin cells of the future medulla and the cells of the fetal cortex derive from the **mesothelium** of the dorsal mesentery mix. These mesothelial cells divide and differentiate into fetal cortical cells. At this point, there is no zonation of any kind, and the fetal cortex and future medulla are one contiguous cluster of cells; identifying individual chromaffin cells during this stage is very difficult. Throughout fetal development, the fetal cortex works together with the placenta as a **fetal-placental unit**. The fetal cortex lacks some of the machinery of the permanent cortex that the placenta has. The placenta lacks some of the machinery of the permanent cortex that the fetal cortex has. Together, they synthesize cortisol, aldosterone, and androgens.

Later in development, more cells from the mesothelium of the posterior abdominal wall arrive and surround the mish-mash that is the fetal cortex—both chromaffin cells and fetal cortex cells. These new cells will eventually become the permanent cortex. At 28 weeks' gestational age, there is only a thin layer of permanent cortex that shows only cells that look like the zona glomerulosa. Seventy percent of the adrenal gland mass is the fetal cortex. Over time, the permanent cortex will replicate, differentiate, and become the three zonae of the permanent cortex, while the fetal cortex fulfills the service of hormone synthesis. The completion of the adrenal glands, with complete zonation, obvious medulla, and complete involution of the fetal cortex, occurs around age 4 months. Until birth, there is no definitive medulla, only scattered chromaffin cells amongst fetal cortex cells. At birth, the fetal cortex undergoes rapid involution, which results in a massive size reduction of the adrenal gland. As a result of this involution, the area occupied by the medulla shrinks, and the chromaffin cells cluster together in the center, giving rise to a distinct medulla.

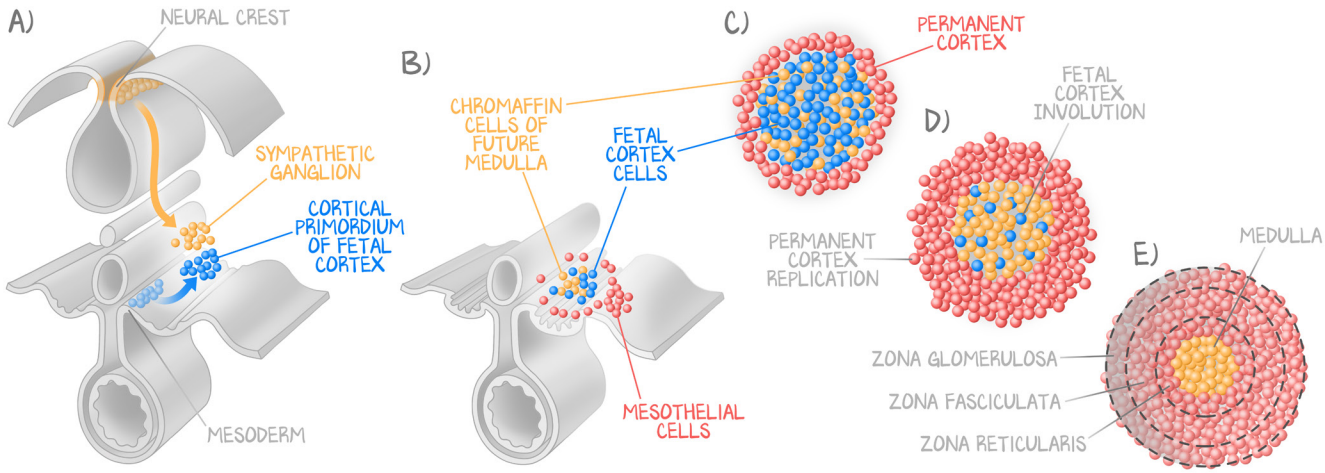

Figure 1.6: Embryogenesis of the Adrenal Gland
(a) The folding of the neural tube and the identification of the neural crest. Mesodermal mesenchyme forms the cortical fetal cortex, whereas the neural crest cells form a typical sympathetic ganglion. (b) Mesothelial cells migrate from the peritoneum to form the adult cortex. The fetal cortex ensures that this sympathetic ganglion will remain an endocrine gland, and not develop axons, as the other ganglia do. (c) Early in development, the permanent cortex contributes very little to adrenal hormone production. (d) Nearing birth, the permanent cortex is dominant over the fetal cortex, which has started undergoing apoptosis. (e) After birth, the fetal cortex is gone, the chromaffin cells in the center of the gland (medulla), and the zona are differentiated from each other.

Steroid Hormone Synthesis and Action

We are going to present you with the pathway for adrenal hormone synthesis. You are not to memorize this pathway. This section allows us to take the shortcuts we do in Adrenal #4: *Adrenal Hyperplasia NOS*.

The *CYP17A1* gene encodes the cytochrome P-450 enzyme **17A1**. This one enzyme coded by one gene performs multiple enzymatic reactions in the synthesis of steroid hormones. **17α-hydroxylase**, **17,20-demolase**, and **17,20-lyase** are all the same enzyme, the same *CYP17A1*. Any of these names may be used, and they are all synonymous. They are named differently based on the enzymatic step they are carrying out, but they are NOT different enzymes.

All steroid hormones are made from **cholesterol**. The first step is the stimulation of the ACTH receptor. Cholesterol is converted by an unnamed enzyme to an unnamed compound. This first unnamed compound can become DHEA, aldosterone, or cortisol if the correct enzymes are present. Aldosterone follows the same enzymatic pathway as cortisol, except that at any time, the aldosterone lineage can

be converted to the cortisol lineage by the enzyme **17α-hydroxylase**. Aldosterone is synthesized only in the zona glomerulosa because the zona glomerulosa cells are the only cells that express **aldosterone synthase**. The fasciculata and reticularis both can make DHEA because the only enzyme that is needed is **17α-hydroxylase** (17α-hydroxylase and 17,20-desmolase are the same enzyme). Without it, the steroid would be trapped in the aldosterone pathway. To become either aldosterone or cortisol, the enzymes **21α-hydroxylase** and **11β-hydroxylase** are required. Without them, only intermediates and DHEA can be made.

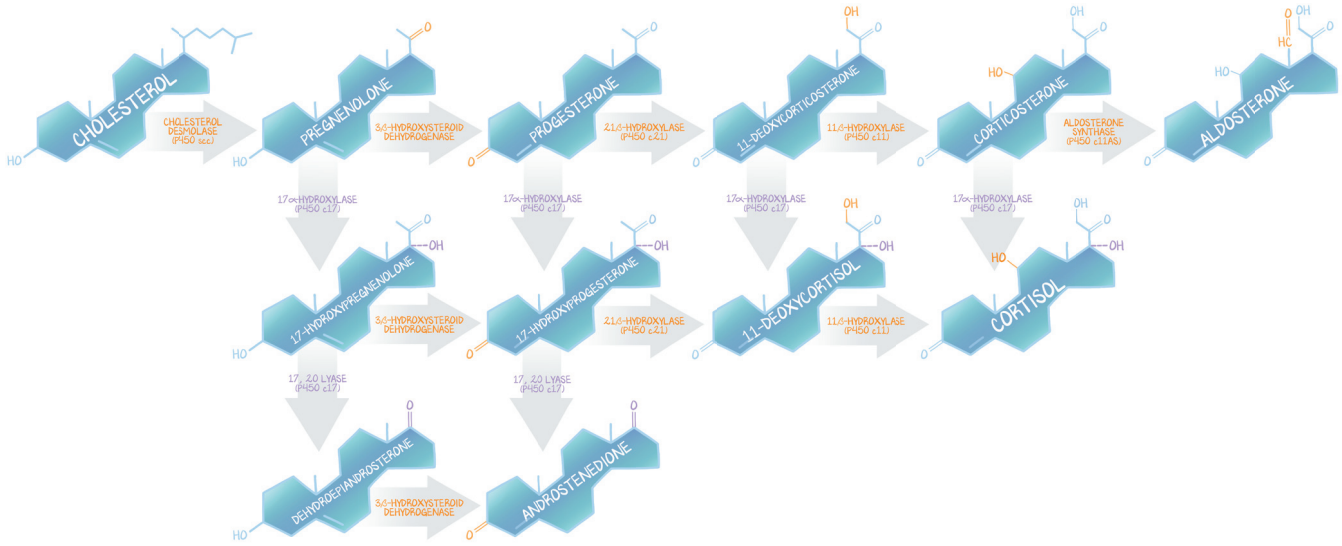

Figure 1.7: Steroid Hormone Synthesis Pathway
This figure is to remind you how difficult it would be to try to learn the pathway in detail. So many enzymes are happening in so many organelles. You will use this as a reference when we simplify this process later. However, do see that these are cholesterol hormones, made from the modification of an unchanging cholesterol backbone, with only the carbons at the edges being modified.

Aldosterone and **cortisol** share the ability to activate mineralocorticoid receptors. Aldosterone does not activate glucocorticoid receptors. Although the mineralocorticoid activity of aldosterone is three thousand times that of cortisol, under normal conditions, the amount of circulating cortisol is about two thousand times that of aldosterone. That makes cortisol two-thirds as good at activating aldosterone receptors (mineralocorticoid receptors). The signal for aldosterone is angiotensin-2; the signal for cortisol is ACTH. Aldosterone's function is only in a few cells, such as the collecting duct of the nephron and the ductal cells of sweat glands. So, to ensure that ACTH-driven cortisol doesn't interfere with angiotensin2-driven aldosterone, the cells that are the target of aldosterone possess an enzyme, 11β-hydroxysteroid dehydrogenase 2 (**11β-HSD2**), which essentially induces an irreversible conversion of cortisol to cortisone, which has no mineralocorticoid activity.

A clinically irrelevant factoid, but one that appears in every textbook and classroom lecture on the subject, is that licorice, in extremely high doses, can inhibit 11β-HSD2, thereby inducing relative hyperaldosteronism due to the abundance of cortisol now acting on aldosterone receptors. To experience symptoms, there would need to be a LOT of licorice ingested over multiple days—something that is unlikely to actually happen.

Finally, some intermediates in the steroid hormone synthesis pathway DO have varying effects on mineralocorticoid and glucocorticoid receptors. We just said cortisone has none. Memorizing the details of the intermediates is not worthwhile—know aldosterone and cortisol and assume the rest are 0. Just be aware that in stripping the process of steroid hormone synthesis down to its bare bones, there is plenty we have intentionally left out.